AMENDMENT TO THE CLAIMS

Claims 1-83. (canceled)

- 84. (previously presented) A method for assaying for modulators of β -secretase activity, comprising:
- (a) contacting a polypeptide with β -secretase APP processing activity with a substrate, both in the presence and in the absence of a putative modulator compound;

wherein said substrate comprises a peptide having an amino acid sequence of at least 6 amino acids, said amino acid sequence including four amino acids defined by formula P_2P_1 - P_1 : P_2 , wherein:

 P_2 comprises an amino acid selected from the group consisting of N, L, K, S, G, T, D, A, Q and E;

 $P_{1} \ comprises \ an \ amino \ acid \ selected \ from \ the \ group \ consisting \ of \ Y, \ L, \ M,$ Nle, F and H;

 $P_{1'}$ comprises an amino acid selected from the group consisting of E, A, D, M, Q, S and G;

 $P_{2'} \ comprises \ an \ amino \ acid \ selected \ from \ the \ group \ consisting \ of \ A, \ V, \ N, \ T,$ L, F and S;

wherein the substrate is cleaved between P_1 and $P_{1'}$ by a human aspartyl protease encoded by the nucleic acid sequence of SEQ ID NO: 1 or SEQ ID NO: 3 (Hu-Asp2); and

wherein said peptide does not comprise the corresponding P₂P₁-P₁·P₂·portion of amino acid sequence depicted in SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28, SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33, SEQ ID NO: 34, SEQ ID NO: 35, SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38, or SEQ ID NO: 39;

(b) measuring cleavage of the substrate peptide in the presence and in the absence of the putative modulator compound; and

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M;

 $P_{3'}$ is E.

- (c) identifying modulators of β -secretase activity from a difference in substrate cleavage in the presence versus in the absence of the putative modulator compound, wherein a modulator that is a β -secretase antagonist reduces such cleavage and a modulator that is a β secretase agonist increases such cleavage.
 - 85. (previously presented) The method of claim 84,

wherein said substrate comprises a peptide having an amino acid sequence of at least 6 amino acids, said amino acid sequence including five amino acids defined by formula $P_2P_1-P_1\cdot P_2\cdot P_3\cdot$ and

wherein $P_{3'}$ comprises an amino acid selected from the group consisting of E, G, F, H, cysteic acid and S.

86. (previously presented) The method of claim 84, wherein the peptide comprises a sequence of amino acids defined by the formula P_2P_1 - P_1 , P_2 - P_3 , wherein

P₂ comprises an amino acid selected from the group consisting of N, S, and D;

P₁ comprises an amino acid selected from the group consisting of Y, F and L;

P_{1'} comprises an amino acid selected from the group consisting of E, A, and D;

P2' comprises an amino acid selected from the group consisting of A and V;

 $P_{3'}$ comprises an amino acid selected from the group consisting of E, G, F, H, cysteic acid and S.

87. (previously presented) The method of claim 84, wherein the peptide comprises a sequence of amino acids defined by the formula P₂P₁-P_{1'}P_{2'}P_{3'}, wherein

P₂ comprises an amino acid selected from the group consisting of S, N, and K;

P₁ comprises an amino acid selected from the group consisting of F, L, Y, and

P_{1'} comprises an amino acid selected from the group consisting of E, D, and A; P_{2'} comprises an amino acid selected from the group consisting of A and V;

- 88. (previously presented) The method of any one of claims 85-87, wherein the peptide comprises a sequence of amino acids defined by the formula $P_3P_2P_1$ - $P_1P_2P_3$, wherein P_3 is an amino acid selected from the group consisting of A, V, I, S, H, Y, T and F.
- 89. (previously presented) The method of claim 88, wherein P₃ comprises an amino acid selected from the group consisting of I or V.
- 90. (previously presented) The method of claim 88, wherein the peptide comprises a sequence of amino acids defined by the formula $P_4P_3P_2P_1-P_1\cdot P_2\cdot P_3\cdot P_4$ wherein P_4 is an amino acid selected from the group consisting of E, G, I, D, T, cysteic acid and S.
- 91. (previously presented) The method of claim 90, wherein the peptide comprises a sequence of amino acids defined by the formula $P_4P_3P_2P_1-P_1P_2P_3P_4$ wherein P_4 is an amino acid selected from the group consisting of F, W, G, A, H, P, G, N, S, and E.
- 92. (previously presented) The method of claim 84, wherein the amino acids at positions P₂, P₁, P₁, P₂ comprise N, F, E and A, respectively.
 - 93. (previously presented) The method of claim 84, wherein:

P₂ is N or S;

P₁ is selected from the group consisting of Y, F, and L;

P_{1'} is selected from the group consisting of E, D, and A; and

P_{2'} is V.

- 94. (previously presented) The method of claim 84, wherein said substrate comprises an amyloid precursor protein (APP) amino acid sequence with a modified β -secretase processing site defined by said formula $P_2P_1-P_1P_2$.
- 95. (previously presented) The method of any one of claims 84-93, wherein said peptide comprises an amino acid sequence having up to 50 amino acids.

- 96. (previously presented) The method of any one of claims 84-94 wherein the peptide further comprises a first label.
- 97. (previously presented) The method of claim 96 wherein the peptide further comprises a second label.
- 98. (previously presented) The method of any one of claims 84-94 wherein the peptide further comprises a detectable label and a quenching moiety, wherein cleavage of the peptide between P_1 and $P_{1'}$ separates the quenching moiety from the label to permit detection of the label.
- 99. (previously presented) The method of claim 85, wherein said cysteic acid comprises a covalently attached label.
- 100. (previously presented) The method of any one of claims 84-94, wherein the rate of cleavage of said peptide by said human aspartyl protease is greater than the rate of cleavage of a polypeptide comprising the human APP β -secretase cleavage sequence: SEVKMDAEFR (SEQ ID NO: 20).
- 101. (previously presented) The method of any one of claims 84-94, wherein the rate of cleavage of said peptide by said human aspartyl protease is greater than the rate of cleavage of a polypeptide comprising the human APP Swedish KM \rightarrow NL mutation, β -secretase cleavage sequence SEVNLDAEFR (SEQ ID NO: 19).
- 102. (previously presented) The method of any one of claims 84-94, wherein the polypepetide with β -secretase APP processing activity comprises an amino acid sequence selected from the group consisting of
 - (a) the amino acid sequence of SEQ ID NO: 2,
- (b) a fragment of the amino acid sequence of SEQ ID NO: 2 that retains β -secretase APP processing activity, wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG,

- (c) an amino acid sequence that is at least 95% identical to (a) or (b), wherein the polypeptide includes the aspartyl protease active site tripeptides DTG and DSG and exhibits β -secretase APP processing activity;
 - (d) the amino acid sequence SEQ ID NO: 4,
- (e) a fragment of the amino acid sequence of SEQ ID NO: 4 that retains β secretase APP processing activity, wherein said fragment includes the aspartyl protease active
 site tripeptides DTG and DSG, and
- (f) an amino acid sequence that is at least 95% identical to (d) or (e), wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG and exhibits β -secretase APP processing activity.
- 103. (previously presented) The method of any one of claims 84-94, wherein the polypeptide with β -secretase APP processing activity comprises an amino acid sequence selected from the group consisting of
 - (a) the amino acid sequence of SEQ ID NO: 2; and
- (b) a fragment of the amino acid sequence of SEQ ID NO: 2 that retains β -secretase APP processing activity, wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG.
- 104. (previously presented)A method according to claim 103, wherein the polypeptide with β -secretase APP processing activity comprises a polypeptide purified and isolated from a cell transformed or transfected with a polynucleotide comprising a nucleotide sequence that encodes the polypeptide.
 - 105. (previously presented) A method according to claim 95,

wherein the substrate is expressed in a cell transformed or transfected with a polynucleotide comprising a nucleotide sequence that encodes the substrate,

wherein the cell expresses the polypeptide with β -secretase APP processing activity;

wherein the contacting comprises growing the cell in the presence and absence of the test agent, and

wherein the measuring step comprises measuring APP processing activity of the cell.

- 106. (previously presented) A method according to claim 105, wherein the contacting comprises administering the test agent to a transgenic non-human mammal that comprises the cell.
- 107. (previously presented) A method according to claim 84, wherein the polypeptide is encoded by a polynucleotide comprising the nucleotide sequence selected from the group consisting of:
 - (a) the nucleotide sequence of SEQ ID NO: 1 or SEQ ID NO; 3,
- (b) a nucleotide sequence that hybridizes under the following stringent hybridization conditions to the complement of SEQ ID NO: 1 or 3:
- (1) hybridization at 42° C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and
- (2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS;

wherein said nucleotide sequence encodes a polypeptide that exhibits β secretase APP processing activity.

108. (previously presented) A method according to claim 84, wherein the substrate comprises a peptide having an amino acid sequence selected from the group consisting of SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 115, SEQ ID NO: 116, SEQ ID NO: 117, SEQ ID NO: 118, SEQ ID NO: 119, SEQ ID NO: 133, SEQ ID NO: 135, SEQ ID NO: 136, SEQ ID NO: 137, SEQ ID NO: 141, SEQ ID NO: 143, SEQ ID NO: 144, SEQ ID NO: 145, SEQ ID NO: 147, SEQ ID NO: 149, SEQ ID NO: 150, SEQ ID NO: 151, SEQ ID NO: 152 and SEQ ID NO: 153.

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109. (canceled)

110. (new) The method of claim 88, wherein the peptide comprises a sequence of amino acids defined by the formula $P_3P_2P_1-P_1\cdot P_2\cdot P_3$, wherein P_3 is V, P_2 is N, P_1 is F, P_1 is E, P_2 is P_3 is P_3 is P_3 is P_4 .

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